

We claim:

- 1. An isolated mammalian c-kit-/c-met- cardiomyocyte precursor cell of muscular origin.
- 2. The cell of claim 1, wherein the cell is a human cell.
- 3. The cell of claim 1, wherein the cell is a mouse cell.
- 4. The cell of claim \(\), wherein the cell is from a fetus, a child, or an adult.
- The cell of claim 1, wherein the cell is in suspension. 5.
- 6. The cell of claim 1, wherein the cell is between about 3 µm and 10 μm in diameter.
- 7. The cell of claim 6, wherein the cell is approximately 4 µm in diameter.
- The cell of claim 1, wherein the cell differentiates into a 8. cardiomyocyte.
- The cell of claim 1, wherein the cell differentiates into a 9. spontaneously beating cardiomyocyte.
- 10. The cell of claim 1, wherein the cell is transduced with a viral vector.



- 11. The cell of claim 1 wherein the viral vector comprises a heterologous nucleic acid.
- 12. The cardiomyocyte of claim 8, wherein the cardiomyocyte expresses GATA-4, troponin-T, L-type calcium channel, or Nkx2.5, or a combination thereof.
- 13. A method of isolating a c-kit-/c-met- cardiomyocyte precursor cell of muscular origin, comprising:

separating cells of less than 40 µm in diameter from a suspension of muscle cells;

culturing the cells in a tissue culture medium on a solid substrate; and

isolating the cells in suspension in the medium; thereby isolating the c-kit-/c-met- cardiomyocyte precursor cell of muscular origin.

14. The method of claim 13, wherein separating cells of less than 40 μm in diameter from a suspension of cells comprises:

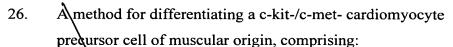
passing the suspension of cells through a first filter with a pore size of about $50-200~\mu m$ to collect a first eluate containing cells of greater than about 50 μm and less than about 200 μm in diameter; and

passing the first eluate through a second filter with a pore size of about 40 µm to collect a second eluate containing cells of less than about 40 µm in diameter.

15. The method of claim 14 wherein the first filter has a pore size of at least 100 μm and the second filter has a pore size of about 40 μm.



- 16. The method of claim 13, wherein the tissue culture medium is a growth medium.
- 17. The method of claim 16, wherein the growth medium is supplemented with a growth factor.
- 18. The method of claim 17, wherein the growth factor is EGF, or bFGF, or a combination thereof.
- 19. The method of claim 18, wherein the growth factor EGF is present at a concentration between about 5 and 50 ng/ml.
- 20. The method of claim\19, wherein the growth factor EGF is present at a concentration between about 5 and 10 ng/ml.
- 21. The method of claim 19, wherein the growth factor EGF is present at a concentration of about 10 ng/ml.
- 22. The method of claim 18, wherein the growth factor bFGF is present at a concentration between about 5 and 50 ng/ml.
- 23. The method of claim 22, wherein the growth factor bFGF is present at a concentration between about 5 and 10 ng/ml.
- 24. The method of claim 22, wherein the growth factor bFGF is present at a concentration of about 10 ng/ml.
- 25. A mammalian c-kit-/c-met- cardiomyocyte precursor cell of muscular origin isolated according to the method of claim 13.



separating cells of less than 40 µm in diameter from a suspension of muscle cells;

culturing the cells in a tissue culture medium in the presence of a growth factor on a solid substrate;

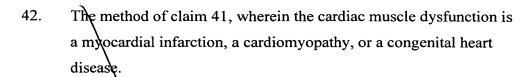
isolating the cells in suspension in the medium; and removing the growth factor, thereby differentiating the c-kit/c-met- cardiomycyte precursor cell of muscular origin into a cardiomycyte.

- 27. The method of claim 28, wherein the cardiomyocyte is spontaneously beating.
- 28. The method of claim 26, wherein the growth factor is EGF, or bFGF, or a combination thereof.
- 29. The method of claim 28, wherein the growth factor EGF is present at a concentration between about 5 and 50 ng/ml.
- 30. The method of claim 29, wherein the growth factor EGF is present at a concentration between about 5 and 10 ng/ml.
- 31. The method of claim 29, wherein the growth factor EGF is present at a concentration of about 10 ng/ml.
- 32. The method of claim 28, wherein the growth factor bFGF is present at a concentration between about 5 and 50 ng/ml.
- 33. The method of claim 32, wherein the growth factor bFGF is present

at a concentration between about 5 and 10 ng/ml.

- 34. The method of claim 32, wherein the growth factor bFGF is present at a concentration of about 10 ng/ml.
- 35. A mammalian cardiomyocyte differentiated from a c-kit-/c-met-cardiomyocyte precursor cell of muscular origin according to the method of claim 26.
- 36. A method of treating a myocardial injury in a subject, comprising administering a therapeutically effective amount of the cell of claim 1, thereby treating the myocardial injury.
- 37. The method of claim 36 wherein the cells are introduced locally into the myocardial injury.
- 38. The method of claim 36, wherein the cells are introduced systemically into the subject.
- 39. The method of claim 38, wherein the cells are introduced intravenously.
- 40. The method of claim 36, wherein the myocardial injury is cardiomyopathy, myocardial infarction or congenital heart disease.
- 41. A method of treating cardiac muscle dysfunction, comprising administering to a subject with cardiac dysfunction a therapeutically effective amount of mammalian c-kit-/c-met- cardiomyocyte precursor cells of muscular origin that differentiate into beating cardiomyocytes.





- 43. A pharmaceutical composition comprising mammalian c-kit-/c-met-cardiomyocyte precursor cells of muscular origin in a pharmaceutically acceptable carrier.
- 44. A method for screening for an agent to determine the effect of the agent on a cardiomyocyte comprising:

providing mammalian c-kit-/c-met- cardiomyocyte precursor cells of muscular origin;

contacting the cells with the agent; and observing the effect of the agent on the cells.

- 45. The method of claim 44, wherein observing the effect comprises determining the effect of the agent on differentiation of the cells.
- 46. The method of claim 45 wherein determination of the effect on differentiation comprises assaying expression of GATA-4, expression of cardiac troponin-T, expression of L-type calcium channel, or expression of Nkx2.5, or a combination thereof.
- 47. The method of claim 45, wherein observing the effect comprises assaying a parameter of cardiomyocyte function of the cells.
- 48. The method of claim 47 wherein the parameter comprises spontaneous beating of the cells.
- 49. A kit for promoting cardiomyocyte differentiation, comprising a container containing a purified population of mammalian c-kit-/c-





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met- cardiomyocyte precursor cells of muscular origin.

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The kit of claim 49, further comprising a container containing a growth factor, a container containing a culture medium, instructions for using the kit, or any combination thereof.

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